Amendments to the Claims

The listing of claims will replace all prior versions, and listings of claims in the application.

1-54. (cancelled)

55. (currently amended) A method for detecting DNA or RNA in a test sample, said method comprising:

(a) hybridizing a single stranded target polynucleotide with an abortive promoter cassette comprising a sequence that hybridizes to the single stranded target polynucleotide, and a region that can be detected by transcription by a polymerase;

(b) incubating said target polynucleotide with an RNA polymerase, an initiator, and a terminator;

(c) synthesizing an oligonucleotide transcript that is complementary to the initiation start site of said abortive promoter cassette by an abortive, reiterative process, wherein said initiator is extended until said terminator is incorporated into said oligonucleotide transcript, thereby synthesizing multiple abortive reiterative oligonucleotide transcripts; and

(d) detecting or quantifying said reiterative oligonucleotide transcripts

wherein the abortive promoter cassette comprises one or more oligonucleotides selected from the group consisting of

(i) one self-complementary contiguous oligonucleotide to which RNA polymerase can bind to form a transcription bubble;

(ii) two partially complementary oligonucleotides that form a transcription bubble from which the initiator and the RNA polymerase synthesize the reiterative transcript; and

(iii) two complementary oligonucleotides that form a transcription bubble in the presence of an RNA polymerase, from which the initiator and the RNA polymerase synthesize the

reiterative transcript.

56. (currently amended) A method for detecting the presence of pathogens in a test sample, said method comprising:

(a) hybridizing a single stranded target pathogen polynucleotide in said test sample with an abortive promoter cassette comprising a region that can be detected by transcription by a polymerase;

(b) incubating said target polynucleotide and an initiator with an RNA polymerase, and a terminator;

(c) synthesizing an oligonucleotide transcript that is complementary to initiation start site of the abortive promoter cassette by an abortive, reiterative process, wherein said initiator is extended until said terminator is incorporated into said oligonucleotides thereby synthesizing multiple abortive reiterative oligonucleotide transcripts; and

(d) determining the presence of a pathogen by detecting or quantifying said reiterative oligonucleotide transcripts synthesized from said test sample

wherein the abortive promoter cassette comprises one or more oligonucleotides selected from the group consisting of

(i) one self-complementary contiguous oligonucleotide to which RNA polymerase can bind to form a transcription bubble;

(ii) two partially complementary oligonucleotides that form a transcription bubble from which the initiator and the RNA polymerase synthesize the reiterative transcript; and

(iii) two complementary oligonucleotides that form a transcription bubble in the presence of an RNA polymerase, from which the initiator and the RNA polymerase synthesize the reiterative transcript.

57. (previously presented) The method of any one of claims 55 or 56, further comprising detecting or quantifying said reiteratively synthesized oligonucleotide transcript by modifying a nucleotide in at least one of the members selected from

the group consisting of said terminator, and said initiator.

58. (original) The method of claim 57, wherein said modifying comprises incorporating a label moiety.

59. (original) The method of claim 58, wherein said label moiety comprises a fluorophore moiety.

60. (original) The method of claim 59, wherein said fluorophore moiety comprises a fluorescent energy donor and a fluorescent energy acceptor wherein said moiety is detected or quantified by fluorescence resonance energy transfer.

61. (previously presented) The method of any one of claims 55 or 56, wherein said polymerase is selected from the group consisting of: a DNA-dependent RNA polymerase, an RNA-dependent RNA polymerase and a modified RNA polymerase, and a primase.

62. (previously presented) The method of claim 61, wherein said polymerase comprises an RNA polymerase derived from one of *E. coli, E. coli* bacteriophage T7, *E. coli* bacteriophage T3, and *S. typhimurium* bacteriophage SP6.

63. (previously presented) The method of any one of claims 55 or 56, wherein said abortive oligonucleotides being synthesized are one of the lengths selected from the group consisting of: about 2 to about 26 nucleotides, about 26 to about 50 nucleotides and about 50 nucleotides to about 100 nucleotides.

64. (previously presented) The method of any one of claims 55 or 56, wherein said terminator comprises a nucleotide analog.

65. (previously presented) The method of claim 55 or 56, wherein said initiator comprises nucleotides selected from the group consisting of: 1-25 nucleotides,

26-50 nucleotides, 51-75 nucleotides, 76-100 nucleotides, 101-125 nucleotides, and 126-150 nucleotides, 151-175 nucleotides, 176-200 nucleotides, 201-225 nucleotides, 226-250 nucleotides, and greater than 250 nucleotides.

66. (previously presented) The method of any one of claims 55 or 56, wherein said single-stranded target polynucleotide is one of DNA and RNA.

67. (previously presented) The method of any one of claims 55 or 56, wherein said inititator is RNA.

68. (previously presented) The method of any one of claims 55 or 56, wherein said initiator comprises nucleotides selected from the group consisting of: 1-25 nucleotides, 25-50 nucleotides, 50-75 nucleotides, 75-100 nucleotides, 100-125 nucleotides, and 125-150 nucleotides, 150-175 nucleotides, 175-200 nucleotides, 200-225 nucleotides, and 225-250 nucleotides.

69. (previously presented) The method of claim 55 or claim 56, wherein said abortive promoter cassette comprises a self-complementary oligonucleotide that forms a single-stranded bubble in the presence of an RNA polymerase, wherein a region of said bubble can be detected by transcription by said polymerase.

70. (previously presented) The method of claim 56, wherein said abortive promoter cassette further comprises an abortive promoter cassette linker which is adapted to hybridize to an end of said target pathogen polynucleotide.

71. (currently amended) A method for detecting pathogens in a test sample, said method comprising:

(a) immobilizing a capture probe designed to hybridize with a single stranded target polynucleotide in said test sample;

(b) hybridizing said capture probe with a test sample that potentially contains said single stranded target polynucleotide;

(c) hybridizing said target polynucleotide in said test sample with an abortive promoter cassette comprising a region that hybridizes to the target pathogen polynucleotide, and a region that can be detected by transcription by a polymerase;

(d) incubating said target polynucleotide with an RNA-polymerase, initiator, and a terminator;

(e) synthesizing an oligonucleotide transcript that is complementary to the transcription initiation start site of said abortive promoter cassette by an abortive, reiterative process, wherein said initiator is extended until said terminator is incorporated into said oligonucleotides thereby synthesizing multiple abortive reiterative oligonucleotide transcripts; and

(f) determining the presence or absence of a pathogen by detecting or quantifying said reiterative oligonucleotide transcripts

wherein the abortive promoter cassette comprises one or more oligonucleotides selected from the group consisting of

(i) one self-complementary contiguous oligonucleotide to which RNA polymerase can bind to form a transcription bubble;

(ii) two partially complementary oligonucleotides that form a transcription bubble from which the initiator and the RNA polymerase synthesize the reiterative transcript; and

(iii) two complementary oligonucleotides that form a transcription bubble in the presence of an RNA polymerase, from which the initiator and the RNA polymerase synthesize the reiterative transcript.

72. (withdrawn) A method for detecting mRNA expression in a test sample, the method comprising:

(a) hybridizing a target mRNA sequence with an abortive promoter cassette comprising a region that can be detected by transcription by a polymerase;

(b) incubating said target mRNA sequence with an RNA-polymerase, an initiator, and a terminator;

(c) synthesizing an oligonucleotide transcript that is complementary to the transcription initiation start site, wherein said initiator is extended until said terminator is incorporated into said oligonucleotide transcript, thereby synthesizing multiple reiterative oligonucleotides; and

(d) determining the presence or absence of the mRNA by detecting or quantifying said reiterative oligonucleotide transcripts.

73. (withdrawn) The method of claim 72, further comprising:

(a) immobilizing a capture probe, wherein said probe hybridizes with a target mRNA sequence;

(b) hybridizing said capture probe with a test sample which potentially contains said target mRNA sequence; and

(c) washing a captured target mRNA sequence to remove unhybridized components of said test sample.

74. (withdrawn) The method of claim 72, wherein modifying further comprises incorporating an independently selected label moiety into at least one of said initiator, said terminator, and said oligonucleotides.

75. (withdrawn) The method of claim 74, wherein said label moiety comprises a fluorophore moiety.

76. (withdrawn) The method of claim 75, wherein detecting comprises detecting by fluorescence resonance energy transfer and said fluorophore moiety comprises one of a fluorescent energy donor and a fluorescent energy acceptor.

77. (withdrawn) The method of claim 72, wherein said polymerase is one of a DNA-dependent RNA polymerase, an RNA-dependent RNA polymerase, an RNA-dependent DNA polymerase, a DNA-dependent DNA polymerase, and a modified polymerase, and a primase.

78. (withdrawn) The method of claim 72, wherein said polymerase comprises an RNA polymerase derived from one of *E. coli*, *E. coli* bacteriophage T7, *E. coli* bacteriophage T3, and *S. typhimurium* bacteriophage SP6.

79. (withdrawn) The method of claim 72, wherein said initiator is one of RNA or DNA.

80. (withdrawn) The method of claim 79, wherein said initiator comprises nucleotides selected from the group consisting of: 1-25 nucleotides, 26-50 nucleotides, 51-75 nucleotides, 76-100 nucleotides, 101-125 nucleotides, 126-150 nucleotides, 151-175 nucleotides, 176-200 nucleotides, 201-225 nucleotides, 226-250 nucleotides, and greater than 250 nucleotides.

81. (withdrawn) The method of claim 72, wherein said abortive oligonucleotides being synthesized are one of the lengths selected from the group consisting of: about 2 to about 26 nucleotides, about 26 to about 50 nucleotides, about 50 nucleotides to about 100 nucleotides, and greater than 100 nucleotides.

82. (withdrawn) The method of claim 72, wherein said abortive promoter cassette comprises a self-complementary oligonucleotide that forms a single-stranded bubble region comprising said target site.

83. (withdrawn) The method of claim 72, wherein said abortive promoter cassette comprises an abortive promoter cassette linker which is adapted to hybridize to a poly-A tail of said target mRNA sequence.

84. (withdrawn) The method of claim 72, wherein said chain terminator is a nucleotide analog.

85-105. (cancelled).

106. (withdrawn) A method for detecting a target protein in a test sample, the method comprising:

(a) covalently attaching the target protein to an abortive promoter cassette by a reactive abortive promoter cassette linker, wherein said abortive promoter cassette comprises a region that can be detected by transcription by a polymerase;

(b) incubating said target protein with an RNA-polymerase, an initiator, and a terminator;

(c) synthesizing an oligonucleotide transcript that is complementary to the transcription initiation start site of the abortive promoter cassette, wherein said initiator is extended until said terminator is incorporated into said oligonucleotide transcript, thereby synthesizing multiple reiterative oligonucleotide transcripts; and

(d) determining the presence or absence of the target protein by detecting or quantifying said reiterative oligonucleotide transcripts.

107. (withdrawn) The method of claim 106 further comprising immobilizing target protein by a target specific probe.

108. (withdrawn) The method of claim 107, wherein said target specific probe is an antibody.

109. (withdrawn) The method of claim 106, wherein said abortive promoter cassette linker is covalently attached to the target protein by thiol-reactive or amine-reactive crosslinking agents.

110. (withdrawn) The method of claim 109 wherein said protein crosslinking agents are selected from the group consisting of: maleamides, iodoacetamides, and disulfides.

111. (withdrawn) The method of claim 106, wherein said target protein is purified or in a cell lysate.

112. (cancelled).

113. (currently amended) A method for detecting pathogens, said method comprising:

(a) obtaining a sample in need of detection of a pathogen

(b) hybridizing a single stranded target pathogen polynucleotide in said sample with an abortive promoter cassette comprising a nucleotide sequence that hybridizes to single stranded target pathogen polynucleotide, and a region that can be detected by transcription by a polymerase;

(c) incubating said target polynucleotide and initiator with an RNA polymerase, and a terminator;

(d) synthesizing an oligonucleotide transcript that is complementary to the initiation start site of the abortive promoter cassette by an abortive, reiterative process, wherein said initiator is extended until said terminator is incorporated into said oligonucleotides thereby synthesizing multiple abortive reiterative oligonucleotide transcripts; and

(e) determining the presence of a pathogen by detecting or quantifying said reiteratively synthesized oligonucleotide transcripts synthesized from said sample wherein the abortive promoter cassette comprises one or more oligonucleotides selected from the group consisting of

(i) one self-complementary contiguous oligonucleotide to which RNA polymerase can bind to form a transcription bubble;

(ii) two partially complementary oligonucleotides that form a transcription bubble from which the initiator and the RNA polymerase synthesize the reiterative transcript; and

(iii) two complementary oligonucleotides that form a transcription bubble in the presence of an RNA polymerase, from which the initiator and the RNA polymerase synthesize the reiterative transcript.

114. (previously presented) The method of claim 113, wherein said method further comprises:

immobilizing an oligonucleotide capture probe which is specific for said target pathogen polynucleotide; and

hybridizing said oligonucleotide capture probe with a denatured DNA sample which potentially contains said target pathogen polynucleotide.

115-129. (cancelled).

130. (previously presented) The method of claim 113, wherein said sample is obtained from the group consisting of: animal, plant or human tissue, blood, saliva, semen, urine, sera, cerebral or spinal fluid, pleural fluid, lymph, sputum, fluid from breast lavage, mucusoal secretions, animal solids, stool, cultures of microorganisms, liquid and solid food and feed-products, waste, cosmetics, air and water.

131. (previously presented) The method of any one of claims 55, 56, 71 or 113, wherein said abortive promoter cassette comprises two partially complementary oligonucleotides that form a bubble region.

132. (previously presented) The method of any one of claims 55, 56, 71 or 113, wherein said abortive promoter cassette comprises two complementary oligonucleotides that form a bubble region in the presence of RNA polymerase.

133. (currently amended) The method of any one of claims 55, 56, 71 or 113, wherein said abortive promoter cassette comprises one <u>self-complementary</u> contiguous oligonucleotide to which RNA polymerase can bind to form a transcription bubble.

134. (original) The method of claim 59 wherein said fluorophore moiety is selected from the group consisting of: 4-acetamido-4'-isothiocyanatostilbene-2,2'disulfonic acid; acridine and derivatives: acridine, acridine isothiocyanate; 5-(2'-aminoethyl) amninonaphthalene-1-sulfonic acid (EDANS); 4-amino-N-[3-vinylsulfonyl) phenyl] naphthalimide-3,5 disulfonate; N-(4-amino-1-naphthyl)maleimide; anthranilamide; BODIPY; Brilliant Yellow; coumarin, and derivatives: coumarin, 7-amino-4-methylcoumarin (AMC, Coumarin 120), 7-amino-4-trifluoromethylcouluarin (Coumaran 151); cyanine dyes; cyanosine; 4',6-diaminidino-2-phenylindole (DAPI); 5', 5"-dibromopyrogallol-sulfonaphthalein (Bromopyrogallol Red); 7-diethylamino-3-(4'-isothiocyanatophenyl)-4-methylcoumarin; diethylenetriamine pentaacetate; 4,4'-diisothiocyanatodihydro-stilbene-2,2'-disulfonic acid; 4,4'-diisothiocyanatostilbene-2,2'-disulfonic acid; 5-[dimethylamino]naphthalene-1-sulfonyl chloride (DNS, dansylchloride); 4-dimethylaminophenylazophenyl-4'-isothiocyanate (DABITC); eosin and derivatives: eosin, eosin isothiocyanate; erythrosin and derivatives: erythrosin B, erythrosin, isothiocyanate; ethidium; fluorescein and derivatives: 5-carboxyfluorescein (FAM),5-(4,6-dichlorotriazin-2-yl)aminofluorescein (DTAF), 2',7'-dimethoxy-4'5'-dichloro-6-carboxyfluorescein (JOE), fluorescein, fluorescein isothiocyanate, QFITC, (XRITC); fluorescamine; IR144; IR1446; Malachite Green isothiocyanate; 4-methylumbelliferoneortho cresolphthalein; nitrotyrosine; pararosaniline; Phenol Red; B-phycoerythrin; o-phthaldialdehyde; pyrene and derivatives: pyrene, pyrene butyrate, succinimidyl 1pyrene; butyrate quantum dots; Reactive Red 4; rhodamine and derivatives: 6-carboxy-X-rhodamine (ROX), 6-carboxyrhodamine (R6G), lissamine rhodamine B, sulfonyl chloride rhodamine (Rhod), rhodamine B, rhodamine 123, rhodamine X isothiocyanate, sulforhodamine B, sulforhodamine 101, sulfonyl chloride derivative of sulforhodamine 101 (Texas Red); N,N,N',N'-tetramethyl-6-carboxyrhodamine (TAMRA); tetramethyl rhodamine; tetramethyl rhodamine isothiocyanate (TRITC); riboflavin; rosolic acid; terbiun chelate derivatives; Cy 3; Cy 5; Cy 5.5; Cy 7; IRD 700; IRD 800; La Jolla Blue; phthalo cyanine; and naphthalo cyanine.

135. (previously presented) The method of any one of claims 55, 56, 71 or 113, wherein said initiator is selected from the group consisting of: nucleosides, nucleoside analogs, nucleotides, and nucleotide analogs.

136. (withdrawn) A method for detecting DNA or RNA in a test sample, said method comprising:

(a) hybridizing a single stranded target polynucleotide with an abortive promoter cassette comprising a sequence that hybridizes to the single stranded target polynucleotide, and a region that can be detected by transcription by a polymerase;

(b) incubating said target polynucleotide with an RNA polymerase and an initiator;

(c) synthesizing an oligonucleotide transcript that is complementary to the initiation start site of said abortive promoter cassette, wherein said initiator is extended until termination occurs through nucleotide deprivation; thereby synthesizing multiple reiterative oligonucleotide transcripts; and

(e) detecting or quantifying said reiterative oligonucleotide transcripts.

137. (withdrawn) The method of claim 136 further comprising:

(a) immobilizing a capture probe designed to hybridize with a target polynucleotide in said test sample;

(b) hybridizing said capture probe with a test sample that potentially contains said target polynucleotide.

138. (previously presented) The method of claims 56, 71 or 113, further comprising incubating said target polynucleotide with additional ribonucleotides.

139. (previously presented) The method of claim 138, wherein said ribonucleotides are modified.

140. (previously presented) The method of claim 139, wherein said modification comprises incorporating a labeling moiety.

141. (withdrawn) The method of claim 136 for detecting the presence of pathogens in a test sample.

142. (previously presented) The method of any one of claims 56, 71 or 113, wherein the pathogen is a virus.

143. (previously presented) The method of any one of claims 56, 71 or 113, wherein the pathogen is bacterial.

144. (previously presented) The method of any one of claims 55, 56, 71 or 113, wherein the target polynucleotide is RNA.

145. (previously presented) The method of claim 144, wherein the RNA is mRNA.

146. (previously presented) The method of claim 144, wherein the RNA polymerase is an RNA-dependent RNA polymerase.

147. (previously presented) The method of claim 146 wherein the RNA-dependent RNA-polymerase is poliovirus RNA polymerase.

148. (previously presented) The method of claim 144, further comprising incubating said RNA with a reverse transcriptase enzyme.